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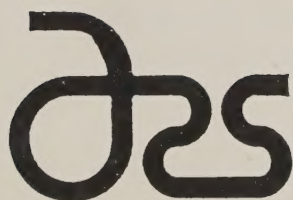
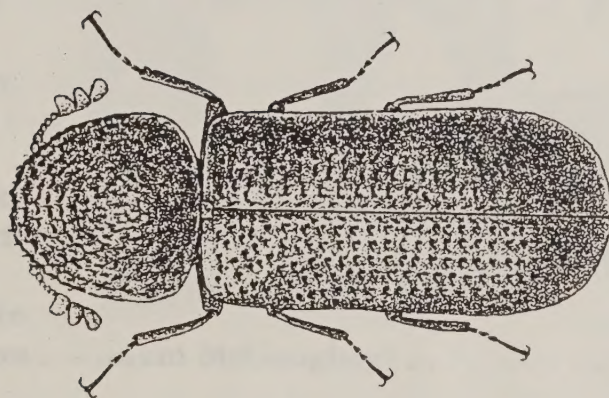
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STORED-PRODUCT INSECTS WORKSHOP

May 4-6, 1992
Savannah, Georgia

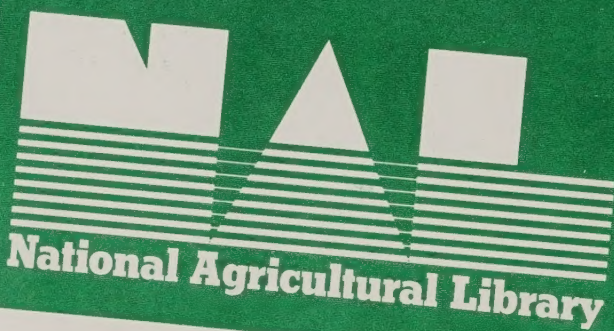
FINAL REPORT



Agricultural
Research
Service

United States Department of Agriculture

**United States
Department of
Agriculture**



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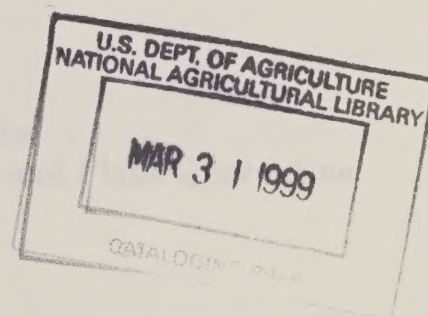
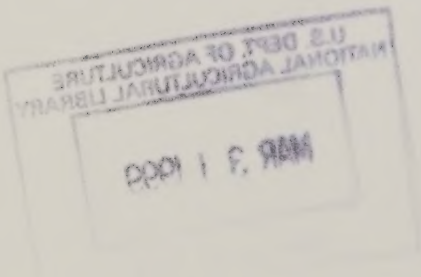


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PREFACE

An Agricultural Research Service (ARS) Workshop on Stored Product Insects was held May 4-6 in Savannah, Georgia. The major objective of this conference was to demonstrate how ARS research projects singularly and collectively form an effective response to specific national problems, and thus conform to the Agency's program plan. Other objectives were 1) define high priority research areas that could be conducted with current resources and 2) define high priority research areas that require new resources. The ARS National Program Staff (NPS) utilizes workshops to help with proper development of research projects and programs to ensure that they are focused on specific national needs. The NPS has the responsibility to prioritize problems, objectives and resource needs for use in guidance of ARS research programs.

In preparation for the workshop, each participant, or group of participants, responsible for a CRIS provided a summary that included objectives, significance, justification(s), constraints, and selected references. This material was provided to all researchers and selected industry participants prior to the meeting. Those documents, along with the following summary report, will provide guidance to not only the NPS but also to the food industry and the involved researchers.

The National Program Staff expresses its gratitude and appreciation to all workshop attendees for their willing participation. The workshop was successful and met the outlined objectives. Special appreciation is extended to the invited speakers: Robert Richardson, Gary Obenauf, Henry Bahn, Thomas Czapla, Michael Fitzner, and Shlomo Navarro, and the invited panel members: Alan Barak (APHIS), David Mueller (Insects Ltd), Jim Rutledge (Lauhoff Grain Company), Terry Pitts (Gustafson, Inc.), Daryl Faustini (Philip Morris USA), Susan McCloud (California Almond Board), John Giler (FGIS), Barrie Kitto (University of Texas), James Touhey (EPA), for their special contributions to the workshop and for their support and interest. The NPS extends a special thanks to the steering committee members: Drs. Richard Arbogast, James Coffelt, William McGaughey, Patrick Vail, and Wendell Burkholder (Chair). The NPS is also grateful to those who were assigned to serve as moderators, organizers, and reporters for the areas of research. Thanks to all participants for having made this workshop a success.

Kenneth W. Vick
National Program Leader
Stored Product Insects and Plant Quarantine

EXECUTIVE SUMMARY

The Agricultural Research Service has been constantly challenged to discover new principles and safer methods of controlling insects in our stored food products. Perhaps now, with declining research funding and greater restrictions on pesticide use, there is the need for improving our priority setting. We need to evaluate the present program, our facilities, and the balance of basic and applied research. More than 30 ARS scientists are now involved in various aspects of stored products insect research at 7 different locations: Beaumont, TX; Gainesville, FL; Fresno, CA; Madison, WI; Manhattan, KS; Oxford, NC; and Savannah, GA. The challenge is how to more effectively manage research and strengthen ARS within the present climate of reduced funding and downsizing.

An ARS workshop on stored product insects was held May-4-5, 1992, in Savannah, Georgia. The aim of this workshop was to summarize the current research program and to relate it to the current national problems such as our concern for developing safe yet effective pest control measures for the food industry that produces, processes, and delivers the food we eat.

The areas of research and the workshop program were arranged in five major areas of emphasis that included:

- (1) Behavior, Ecology and Computer Modeling
- (2) Biological Control
- (3) Insect Pest Management Systems (IPM), Sampling and Detection
- (4) Chemical and Physical Control Methods
- (5) Physiology, Biochemistry, Molecular Biology and Genetics

In the following report each of these areas is presented by 1) current research and 2) research priorities.

In the closing discussion by industry and action agency representatives on "Problems and Research priorities," there were many high priority research needs presented. The industry representatives emphasized that we need a better understanding of the post-harvest pests, control methods, and packaging barriers.

They are asking for a better understanding of resistance to chemicals and more research on substitutes for fumigants. They are asking, "What is the role of pesticides in IPM programs?"

Industry recognizes the importance of sanitation for insect control but they need better detection methods and behavioral studies in commercial settings. They want to know "where do the insects come from." They expect improvements in the current detection and monitoring methods. Industry recognizes the complex nature of the food distribution channels where cross contamination by insects among food products take place. Research on chemical barriers for packaging would be valuable.

The action agencies expressed concern about losing methyl bromide. They are interested in more research on specialized insect detection tests to be used in the field that are fast, cheap, and repeat well along the food channel. They need to know how newly developed techniques correlate to current detection practices.

PHYSIOLOGY, BIOCHEMISTRY, MOLECULAR BIOLOGY AND GENETICS RESEARCH

INTRODUCTION

The number of conventional pesticides available for use in controlling stored product insect pests is declining steadily. The loss of these pesticides is due to environmental and health concerns, acquired pest resistance, and increased costs of development and reregistration. The development of alternative pest management strategies that are environmentally compatible and have reduced health risks is a necessity if the agricultural industry in the United States is to maintain its preeminent position in world commerce. New management systems for stored product pests will incorporate a variety of cultural, physical, biological and chemical control technologies.

Replacement control methods require an understanding of the basic biology of the target pests so that selective disruption of physiological and genetic processes occurs. The goal of research on the physiology, biochemistry, molecular biology and genetics of stored product insects is to provide greater knowledge about insect growth and development that will facilitate the development of these alternative control technologies.

ARS research on the physiology, biochemistry, molecular biology and genetics of stored product insects has focused primarily on the following biological processes because they offer the greatest potential as targets for insect control methods: (1) reproduction, (2) genetic regulation, (3) regulation of growth and development, (4) regulation of digestive processes, (5) regulation of cuticle formation; and (6) pest resistance mechanisms. Technologies based on research findings in these areas will have a high probability of being adapted for commercial use by the agricultural biotechnology industry.

CURRENT RESEARCH

Reproduction. Two areas of reproduction are being examined in moths, the production and development of germ cells, and the physiology and endocrinology of egg production. A unique protein that is produced in germ cells of both male and female moths has been identified and is being characterized. The developmental sequence of germ cell division, follicle formation and oocyte maturation in females is being determined in order to identify hormonal components that regulate egg production.

Yolk proteins packaged into eggs of moths have been identified and the physiology of production and uptake is being determined. Hormones, called ecdysteroids, regulate the major events of egg development. Insect growth regulators (IGRs) that mimic these hormones can be used to disrupt reproduction in stored product insects.

Genetic Regulation. A novel class of parasitic genes that are widespread in natural populations of a stored product insect has been discovered and is being characterized. The genes are self-perpetuating and invasive, and are potentially useful as vectors for modification of pest populations and as tools for population monitoring. They are lethal to non-carrier larvae by maternal action.

A transposable genetic element (TE) in a stored product insect has been cloned and is being characterized. This work will facilitate use of TE-based technology for genetic manipulation of pest species, including the cloning, handling and tailoring of insect genes relevant to growth and development, life cycle disruption, crop virulence and pesticide/pathogen resistance.

Genetic engineering of the genes for germ cell and yolk proteins is being used to develop methods for direct sterilization of stored product insect pests.

A cDNA for insect chitinase was cloned and is being genetically engineered into plants and biological control agents to enhance resistance and pathogenicity to insect pests.

Regulation of Growth and Development. Applications of IGRs for control of insect pests is increasing, but their mode of action is largely unknown. Hormonal regulation of the metabolism of developmental proteins (e.g. storage proteins) is being studied to determine the mechanism of action of IGRs. Storage proteins have been identified and characterized; the genes for several of these proteins have been sequenced. One of the developmental proteins functions in the storage and metabolism of a growth factor during metamorphosis.

A new class of hormones, the eicosanoids, are being characterized in stored product insects. Inhibitors of eicosanoid metabolism were shown to disrupt renal water secretion, the immune response, and body temperature maintenance. IGR's will be designed to disrupt eicosanoid metabolism.

Regulation of Digestive Processes. Digestive enzymes of stored product insects, their naturally occurring proteinaceous inhibitors, and vitamin binding proteins are being studied to identify genes for proteins that might be manipulated for host plant resistance to insects. Target proteinases and carbohydrases from gut tissues of stored product insect pests were purified and characterized.

cDNAs encoding naturally occurring inhibitors of these enzymes are being cloned, sequenced and the recombinant inhibitors evaluated as dietary inhibitors of larval growth. Mixtures of the inhibitors were substantially more active than were the individual inhibitors. Two vitamin-binding proteins were toxic when fed to stored-product insect pests. The enzyme inhibitors and antinutritional proteins are being developed as host plant resistance factors.

Regulation of Cuticle Formation. Insect cuticle structure and metabolism are being studied to identify a) genes for proteins that would help biological control agents to penetrate the insect's protective exoskeleton and gut lining, and b) inhibitors of cuticle physiology. The structures of several crosslinks that form between protein and chitin during cuticle sclerotization are being elucidated. Chitinolytic and proteolytic enzymes from insect molting fluid are being isolated and characterized. Molting enzyme genes are being cloned.

The production of chitin in whole insects and in imaginal discs and cell lines from stored product insects is being used as a model system for studying the mechanism of action of hormones and IGRs. Chitin synthesis inhibitors have been identified and are being tested for efficacy in controlling stored product insects.

Pest Resistance Mechanisms. There is a need to improve the effectiveness of microbial insecticides such as Bacillus thuringiensis (Bt) towards stored product insects. To determine the mechanisms of action and insect resistance, the interaction between individual toxins with insect midgut tissues is being examined. Specific receptors on the midgut epithelial membrane for B+ toxins were discovered and are being characterized. Resistance in certain strains is caused by the loss of these receptors. Host gut pH and proteinase activity had little or no effect on host specificity and susceptibility. The molecular basis for differential susceptibility of Coleoptera and Lepidoptera to B+ toxins is being investigated. Bt variants that circumvent existing resistance mechanisms are being sought.

RESEARCH PRIORITIES

- Develop knowledge of basic biology, biochemistry and genetics of insect pests and their predators, parasites, pathogens and symbionts.
- Identify insect-specific physiological and genetic target sites for insect growth regulators.
- Identify biologically-based materials that disrupt the life cycle of insects.
- Identify genes for insect-specific inhibitors and toxins that can be manipulated for insect pest management.
- Determine basic mechanisms whereby insect growth regulators and biological control agents disrupt insect development.
- Determine potential for use of parasitic DNA and transposable elements as probes and suppressors of insect populations.
- Develop DNA probes for studying and identifying stored product insects, their population structure and sources of infestation.
- Determine mechanisms regulating reproductive physiology, germ cell production, digestive physiology, cuticle physiology, diuresis, and immunity.
- Determine basic mechanisms whereby stored product insects have developed resistance to existing control methods.
- Determine potential for stored-product insects to develop physiological resistance mechanisms to new control agents.
- Determine physiological roles of symbiotic and other microorganisms in insect biology and pathology.
- Determine effects of biological, physical, and chemical control methods on the physiology of stored product insects.
- Determine effects of diet on insect growth, behavior and management practices.

BEHAVIOR, ECOLOGY, AND COMPUTER MODELING

INTRODUCTION

Ecology and behavior form the basis for knowledgeable pest control decisions. Computer models provide a tool for synthesizing results of ecological research studies into a form usable by other researchers and pest control personnel. Much of the older ecological data are of limited use in field applications because the data were collected under conditions that are not typical of actual storage situations. A recent goal of ARS ecological programs on stored-product insects has been to develop computer simulation models using existing data, determine the validity of those models and the shortcomings in the data sets, collect data required to improve the validity of the models, and to use those models to optimize pest control strategies. Thus, these models are now being used cooperatively with other ARS and non-ARS scientists working with biological, cultural, and chemical control technologies to develop a strong ecological basis for our pest management strategies. Ecological studies are being conducted at all ARS stored-product laboratories.

CURRENT RESEARCH

Information on behavior of stored-product insects is generally found within studies on chemical ecology, population biology, and environmental effects. Predominant among past accomplishments were the identifications of sex and aggregation pheromones for nearly all of the major stored-product insects. Sex pheromones from eight species of moths and sex or aggregation pheromones from 26 species of beetles have been identified over the past 25 years. Several of these pheromones have been exploited commercially for monitoring and detection, and the study of others has provided valuable behavioral information on mating and aggregation. Some work has been done on behavioral responses of insects to pheromone-baited traps of various designs. Semiochemicals from cracked grain, oils, and other food products have been found to attract insects outright or enhance attraction to pheromones. Fungal volatiles have been studied that attract insects, while others are repellent. Limited work has been done on kairomones that are used by parasitoids in their orientation to hosts. Other non-semiochemical work has investigated the influence of temperature and humidity on movement of grain and flour beetles. The effects of particle size on reproductive behavior and movement by certain pests and natural enemies has also been studied. Some research focused on the functional responses of natural enemies to host densities. It is apparent that basic behavioral studies, coupled with studies of ecology and physiology need to be conducted in a comprehensive manner in the future.

Insect ecology is the study of the factors regulating insect distribution and abundance. Pest management is applied ecology. With a knowledge of the factors regulating insect distribution and abundance, we can forecast where and when control will be needed. Extensive laboratory studies have been conducted on the effects of temperature, moisture, and commodity on insect behavior and life history. These laboratory studies have been used to increase our understanding of the behavior and life histories of insects in storage facilities by serving as the basis for the development of models that predict the population growth rates of insect pests. We have studied the seasonal changes in insect numbers and distribution in a number of raw commodities including citrus pulp, peanuts, cowpeas, wheat, and corn. Temperature and moisture are the most important factors regulating insect population growth, but parasitoids, pathogens, photoperiod, diapause, and the type of commodity also have been found to be important. Our studies provide insight into how these factors determine insect distribution and abundance under actual storage conditions and have been used to test the accuracy of the predictions of our population growth models. The storage ecosystem is not a closed system. Using pheromone traps, stored-product insects have been shown to be common outside storage facilities. Seasonal patterns of insect flight outside grain bins on farms have been studied. Insects have been shown to infest wheat after storage. Prior to harvest, cowpeas and corn have been shown to become infested by stored-product insects. Ultimately, we need to treat the entire marketing system as a single continuous ecosystem, because insects are moved through the marketing system along with commodities. Studies to date have focused upon farm-stored grain. We are reaching a point at which we have the knowledge to plan and conduct comprehensive studies of insect populations as they are moved through the marketing system with the commodities.

Computer simulation models have been developed that synthesize data from studies on the behavior, ecology, and control of insect pests and data from studies to characterize the stored-product environment. The resulting models allow rapid and thorough examination of various control strategies, or combinations of control strategies, to optimize pest management in stored products. The models allow simulation of experiments that would normally take years, and much money, to conduct. Computer simulation models have been developed for the five major pests of wheat and for four of the major pests of corn. The models for the wheat pests have been coupled with a model developed in Canada for simulating temperature changes in a bin of grain. Additionally, population dynamics models

have been developed for parasitoids of the maize weevil and rusty grain beetle, and are being used as tools in developing biological control systems. A more general model has been developed to predict the movement of grain and seasonal changes in lesser grain borer population levels throughout the wheat marketing system.

RESEARCH PRIORITIES

- Study parasitoid/host and predator/prey interactions to describe and model functional responses to pest densities under natural conditions and determine the roles of chemical, physical, and experiential factors in host location behavior of natural enemies.
- Develop a thorough understanding of the roles played by semiochemicals in the natural context, including pheromones produced by both males and females, and odors produced by food substrates; and chemically identify unknown semiochemicals.
- Describe behaviors associated with movement of insects into and away from stored-products during infestation, dispersal, and control procedures, and study the behaviors of storage pests in wild, non-storage habitats.
- Quantify the effects of various factors on insect survival in stored-products, especially the effects of high and low temperatures.
- Study the dispersal behavior of insects, and provide explanations for their distributions in bulk and packaged commodities, and around storage and processing facilities.
- Design and conduct comprehensive studies of insect populations as they are moved throughout the marketing system with commodities.
- Determine applicability of population models to other strains of the insects in the U.S. and to other types or varieties of grain.
- Quantify the costs and benefits of insects and control measures in stored products, and use the resulting data to develop an economic basis for pest control in stored-products.

- Quantify the effects of temperature and moisture on degradation of protectant chemicals; and quantify the effects of temperature, moisture, concentration of protectant chemical or fumigant, and time of exposure on survival of all stages of pests and beneficial insects in stored-products.
- Use results of simulation studies to optimize grain storage structures, including aeration-fan controllers that base decisions on pest management rules and designing new storage structures that have built-in environmental monitoring equipment that feed data directly to computer models.

IPM SYSTEMS, SAMPLING, AND DETECTION

INTRODUCTION

Much has been accomplished towards developing IPM programs for the stored product industry. New traps and pheromones have been devised, acoustical and immunological detection methods have been developed, and an expert system for grain management was created and is being tested in a pilot project.

CURRENT RESEARCH

Expert Systems and IPM. ARS scientists have developed and validated models for all five major insect pests of stored wheat. The models have routines that predict the effects of aeration, immigration, fumigation, and protectants on insect growth. The insect simulation models have been coupled with a bin environment model that predicts seasonal changes in temperature and moisture for different locations within a grain bin. Population dynamics models are being developed for the hymenopteran parasites that attack stored grain insect pests. Information from simulation studies with the models is being used to develop optimal IPM strategies for stored grain management.

The models are also used in an expert system that is designed for management of insect pests of stored grain. Stored Grain Advisor (SGA) runs on either Macintosh or Microsoft Windows compatible computers. SGA uses simulation models to predict the likelihood that grain will become infested, based on initial grain moisture, length of storage, bin sanitation, grain protectant and aeration. This expert system also provides a graphical key to identify stored-grain insects, and information on when and how to sample the grain for insects. It dynamically changes action thresholds for probe trap catch using statistical equations to correct for duration of sampling and species differences in probe-trap-catch susceptibility.

A three year pilot study to validate the expert system on farm-stored grain was initiated. Analysis of the first year of data indicated that the system accurately predicted when, and under what conditions insects reached damaging levels. The expert system is being adapted to predict infestations in various segments of the wheat marketing system.

Sampling and Trapping. Good sampling methods are the key to effective integrated pest management programs. Sampling studies on stored product pests have helped to define the methods needed to collect ecological data, to validate population growth models, and to determine whether control is needed or has been effective. Statistical equations have been developed that predict the sampling effort required to detect insects over a range of insect densities. These equations are applicable to many different storage situations.

Several new trap designs have been developed for stored-product beetles that take advantage of the differences in insect behavior between species. A plastic probe trap developed for the early detection and monitoring of insects in stored grain is widely used by farmers and others in the grain industry.

Another method of insect sampling that has been developed by ARS scientists in collaborative studies is the ELISA (Enzyme-Linked Immunosorbent Assay) that offers much promise for use in some grain samples and milled cereal products, and a replacement for labor-intensive insect fragment counts. This method, now commercially available, provides a means of quantifying contamination by living and dead insects by measuring the amount of the insect muscle protein myosin in a sample.

The use of insect pheromones and other semiochemicals constitute an important tool in the detection and surveillance of insect pests of stored products. ARS scientists have played a major role in this area, conducting laboratory and field studies to identify the most effective blends, purity and dosages of semiochemicals that govern the behavior of many pests, including the Indianmeal moth, almond moth, cigarette beetle, lesser grain borer, warehouse beetle and grain weevil. Pheromones have been used to control pests in bagged and harvested walnuts, and along with grain probe traps have been used to monitor various pests in and around food processing facilities and warehouses. Optimum dosages and blends of pheromonal components that are commercially available for the Indianmeal moth and the almond moth have been identified and the influence of population density upon pheromone trap effectiveness has been established for the almond moth.

Acoustic Detection. Recent technological advances have enabled ARS scientists to develop acoustic and other electronic methods for the detection and monitoring of adults and larvae of the major insect pests of stored products (including

internal feeders). Studies have shown that insect population density is highly correlated with the number of sounds detected, and that detection distances and temporal differences vary with the species and life stage. These findings have led to the development of an automated acoustic detection system for the continuous monitoring of insect generated sounds in stored grain. This technology is presently being evaluated in a pilot study using farm stored grain.

A desktop unit acoustic detection system (ALFID: Acoustic Location Fixing Insect Detector) for quantifying the number of insects in a grain sample is being developed for use when grain is inspected in marketing channels. The method significantly improves current technology by detecting internally-feeding larvae, which are not detected by conventional sampling techniques. More sensitive detectors and new data analysis methods are being examined to further enhance this new technology prior to field evaluation of the system with industry cooperators .

The widespread acceptance and use of grain probe traps stimulated the development of a prototype design (EPIC: Electronic Probe Insect Counter) that modified the existing commercial trap to enable electronic counting of insects as they fall into a probe trap. Initial tests of the prototype have shown that it provide greater than 95% accuracy in counting adults of the major grain insect pests. Automation of trap data readings eliminates the need for manual inspection and counting. It provides continuous and instantaneous monitoring of trap captures either by a simple LED control box located outside of a storage bin, or through a data transmission system (SMARTS: Serial Multiplexing Addressable Remote Transmission System), where data from many probe traps is transmitted to a central computer. This system is capable of either manual or automatic sequencing through any programmed order of remote locations. A second version of this system is in development that will be capable of identifying species of the captured insects.

RESEARCH PRIORITIES

Expert Systems and IPM

- Develop optimal, integrated pest management programs for various commodities and locations in the marketing system, using modeling and expert systems. These systems will integrate chemical, biological and physical controls using biorational strategies.

- Conduct simulation studies with the bin environment-insect growth model to investigate the effects of latitude, bin size, beneficial insects, and aeration. Incorporate the new model into the expert system.
- Incorporate economic considerations into the models and expert system.
- Adapt the expert system to include other commodities and segments of the grain marketing system: elevators, warehouses and the milling industry.
- Interface the acoustical detection system with the expert system—this system would be targeted primarily for commercial grain storage.

Sampling and Trapping:

- Isolate, identify and synthesize minor pheromone components so that optimum semiochemical lures may be developed.
- Conduct behavioral studies in the field and laboratory to understand how insects orient to semiochemical sources in the relatively still air of the stored product environment.
- Develop methods to characterize the relationship between trap capture and actual population levels, and incorporate this information into IPM programs.
- Study the interaction between the pheromones of stored-product insects and beneficial insects.
- Improve trap/lure combinations and placement of semiochemical-baited traps in the storage environment to optimize trap catch and enhance interpretation of data: identify the potential for direct use of semiochemicals to control pest populations.
- Improve and evaluate the ELISA-based method for insect or insect fragment quantification for rapid field and analysis.

Acoustic Detection:

- Evaluate alternative acoustic sensors, develop more sophisticated detection methods, and complete construction of a sound attenuation chamber to improve the accuracy of ALFID.

- Conduct fundamental biological studies to identify conditions for maximizing sounds produced by insect feeding and movement in bulk and processed stored commodities. Define potential for using EPIC to identify temporal and other variations in insect behavior.
- Use automatic acoustical detection system to collect ecological data from grain storage bins, and scale-up acoustical detection system to collect data from grain elevators.

CHEMICAL AND PHYSICAL CONTROL METHODS

INTRODUCTION

Historically, physical and chemical control methods have been the mainstay of pest control in postharvest technology. In spite of recent advances in alternative technologies, they remain an integral part of IPM programs. Much research has been done in ARS laboratories on chemical and physical control strategies but a great many unanswered questions remain regarding their use and implementation.

CURRENT RESEARCH

Physical Control. Because of the concern about food safety as related to residues and the possible impact of currently used fumigants on the environment, it is important for ARS to research alternatives to chemical control methods. All of these treatments provide protection against insects without leaving undesirable residues and do not contaminate the environment. However, in order to be used on a commercial scale, they must be placed into the storage/processing systems with minimum intrusion, have little or no effects on quality, and be economically feasible. Physical treatments include high temperatures, low temperature, impact, removal, infra-red, microwaves, gamma or ionizing radiation, dehydration, modified atmospheres, and ultrasonics.

Application of heat, low temperatures, physical impact or removal have been used to control insects either actively or passively since commodities have been produced in adequate quantities to require storage. The techniques have developed from the very primitive to highly sophisticated. Various schemes have been devised for the application of either cold or high temperatures. Some depend on generated temperature applications, others depend on the use or application of natural sources, i.e., cold night air. Currently basic and applied research on the use of high and low temperature applications is being conducted at the ARS laboratory in Fresno, California.

In many cases, previous research has been conducted on short term treatments analogous to the use of fumigants. However, like fumigants, the commodity is susceptible to reinvasion after application. In some cases, using physical methods for long term storage has been devised. The application of cold (night air) or high ambient temperature to stored commodities can provide long term storage protection, but is obviously very dependent upon environmental conditions. In addition, there is application potential using fluidized beds or flash cooling or heating methods. In some cases heat is routinely applied (dehydration processes) to the commodity but is

not applied in a uniform manner. Unlike the high value, low volume dried fruit and nut industry, the sheer volume of stored grains may make application of these technologies difficult. Flash heating or cooling methods need further investigation.

Radiation as a disinfestation method was studied intensively during the 1960s and 70s on a worldwide scale. It is presently approved for use on food commodities when applied at 100 krad or less. Few commercial radiation facilities for agricultural products are in use. The use of high velocities to kill insects by impact has also been investigated but is not now in use. Removal of insects infesting commodities is used on a limited scale, mostly by developing countries. Other methods include grain cleansing and trapping. Microwaves and ultrasonics have been investigated in the laboratory, but only limited commercial use is known.

Chemical Control. A significant trend has been an increase in the number of insect growth regulators (IGRs), biological insecticides and the like, for testing. Also, there has been an increase in the number of pyrethroids available for testing. Presently evaluation of these materials is being carried out under a variety of temperature and humidity conditions so that the data are more amenable to use in modeling experiments.

The Savannah laboratory tested more than 350 compounds as repellents in basic and follow-up screen tests. Nineteen of these were classed as effective. Some 35 different insecticides have been evaluated in several laboratories as protectants for rice, corn, wheat, and peanuts. Those showing promise in various types of tests for use against one or more species of pests were pirimiphos-methyl, chlorpyrifos-methyl, fenoxycarb, tralomethrin, fenvalerate, trimethacarb, methoprene, insecto and deltamethrin. Some studies on combination treatments have also been done at Savannah. Relative biological activities of some of these materials have been tested on various substrates and substrate treatments designed to prolong or extend the active life of the pesticide. They have also been tested for efficacy in relation to temperature and moisture content of the commodity. Also, several insecticides have been evaluated as aerosols on bulk and packaged commodities and on woolen fabrics. These include cyfluthrin, prallethrin, permethrin, safrotin, and S-fenvalerate. Cyfluthrin and S-fenvalerate were more effective than dichlorvos in bulk and packaged commodities; permethrin was effective in long-term protection of woolens. In addition to the basic efficacy data in support of registrations and labeling, residue analyses methodology has been

developed. At Savannah, a total of 52 analytical insecticide residue methods have been modified and/or adapted and applied to the residue analysis of 18 different pesticides on 27 different commodities and food fractions.

Research on fumigation and controlled atmospheres has been done at several locations. With the potential loss of methyl bromide as a commodity fumigant, emphasis has been placed on finding alternatives. In the past 5 years only one compound, acrolein, has shown any potential as a fumigant.

Fumigation techniques which improve the efficacy or distribution of fumigant in commodity is another area of importance for assuring the continued efficacious use of these fumigants. In-transit fumigation of grain using phosphine has been researched and the technique of recirculation of phosphine accepted as the method of choice in most situations because it distributes the gas more quickly and uniformly in large bulk masses. Another technique which holds promise is the use of 5% CO₂ with phosphine. By addition of low concentrations of carbon dioxide, the rate of phosphine penetration into bulk and bagged commodities can be increased without using piping or fans for recirculation.

Recent research at Oxford has emphasized limiting the peak emissions of phosphine which occur when tobacco warehouses under fumigation are aerated. New laws in North Carolina require that concentrations of phosphine at the property boundaries not exceed 0.1 ppm. Tests showed that opening only the vents on the warehouses the first day of aeration reduced peak emissions of phosphine by 81%. This information along with sorption/desorption studies of phosphine on tobacco in hogsheads is being utilized to refine a model which will predict conditions and timing for fumigation and aeration.

Because of the emphasis on non-residue producing chemicals for use in protecting agricultural products, controlled atmospheres appear to provide promise as control agents. Recent studies on moth species which attack nuts and stored products have shown that either low O₂ or high CO₂ concentrations are efficacious. Insects are killed more quickly by CO₂ in low relative humidity situations and generally CO₂ concentrations above 40% will kill insects more quickly than low O₂ atmospheres. Conventional fumigants are more toxic to larvae than to eggs and pupae, while low O₂ atmospheres are more toxic to eggs and pupae. Combinations of controlled atmospheres with heat and cold may prove promising for control of insects on grain, dried fruit and nuts.

Pesticide Resistance. This phenomenon has been a consideration in pest control technology for as long as there have been pest control agents. Through the years resistance has increased in intensity and variety and there is every reason to expect that this trend will continue. Resistance surveys from various commodities in Savannah, Manhattan, Oxford, and Beaumont, Texas have shown that resistance to insecticides is pervasive among the major pest species. Control failures with malathion have precluded its use in stored peanuts. Dichlorvos resistance has increased and is inducing cross-resistance to other OPs in field strains. Because of resistance, reldan was shown to be ineffective against lesser grain borer in wheat after being used only 1 year. Reldan resistance has also been demonstrated in lesser grain borer from rice. Resistance to Bacillus thuringiensis has been reported in the Indianmeal moth. Of great concern is the development of resistance to phosphine. High levels have been found in the almond moth, Indianmeal moth, red flour beetle, lesser grain borer, and cigarette beetle. To date, however, control failures with phosphine have been associated with improper fumigation practices rather than with resistance.

Physiological studies have shown that malathion resistance in almond moth and Indianmeal moth results from a highly specific malathionase with a half-life of 3 hours. Also, research has been done on comparing the fitness of the resistant and susceptible genotypes. In an effort to manage resistance, laboratory-collected data on resistance must be correlated with control failures in order to find a test statistic which can predict the success of a particular insecticide against field strains of resistant pests.

RESEARCH PRIORITIES

- More research is needed on the basic biological effects of treatments on the target insect(s), particularly the Coleoptera species.
- Physiological bases (active sites) of physical treatments and controlled atmospheres need to be determined to make these treatments more effective.
- Physical treatments need to be combined with other control methods to provide highly effective, long-term control systems.
- Engineering is needed to provide the most efficient use of alternative treatments such as controlled atmospheres and temperature applications.
- Identify and evaluate biological and environmental factors that affect degradation and efficacy of chemicals, physical treatments, and controlled atmospheres.
- Optimize physical controls such as aeration and grain drying for incorporation into an integrated pest management program.

- Develop methods for effective utilization of insect traps and correlation of trap catch to economic thresholds.
- Develop or adapt methods for residue analyses.
- Investigate the potential of new compounds to be used as fumigants.
- Study the penetrations of gases through packaging materials.
- Investigate the efficacy and usefulness of combining inert gases with fumigants.
- Conduct research on the fate of fumigants in the atmosphere.
- Develop fumigation and controlled atmosphere techniques which can be modeled for IPM programs.
- Conduct research on the use of phosphine and insect pathogens to disinfest and prevent reinfestations of commodities.
- Develop improved methods for sealing facilities for fumigation.
- Develop methods to recapture/recycle fumigants following fumigation.
- Conduct research on the tolerance of diapausing stages of post harvest insects to chemical, physical and controlled atmosphere treatments.
- Develop resistance management program by monitoring field populations for pesticide resistance, developing new methods for detecting and assessing resistance, and determining mechanisms responsible for resistance.
- Investigate the use of insect detection methods such as imaging with near-infrared or X-ray systems and acoustical sensors in sorting systems that eliminate commodity defects.

Biological Control Summary

INTRODUCTION

Research has progressed on many fronts bringing us to the point where a variety of biological control agents (parasites, predators, and pathogens) are ready to test in field-scale experiments. Two actions by the Environmental Protection Agency (registration of Bacillus thuringiensis and exemption of parasitoids and predators from tolerance requirements) make these control agents available for commercial use in stored grains and food storage warehouses.

CURRENT RESEARCH

Parasitoids and predators. A variety of basic laboratory life history and behavioral studies have been reported on several important insect parasitoids (particularly Anisopteromalus calandrae, Bracon hebetor, Choetospila elegans, Cephalonomia waterstoni, Laelius pedatus, Lariophagus distinguendus, Venturia canescens, Trichogramma evanescens, T. pretiosum, and Pteromalus cerealellae), insect predators (Xylocoris flavipes, X. sordidus, Dufouriellus ater, and Lyctocoris campestris) and a predaceous mite (Pyemotes tritici). These attack important insect pests (including lesser grain borer, granary weevil, rice weevil, maize weevil, Indianmeal moth, almond moth, Angoumois grain moth, dermestid beetles, flour beetles, and sawtoothed, merchant, flat, and rusty grain beetles) that occur in bulk-stored wheat, corn, rice, and peanuts and in warehouses.

Kairomones (pest odors attractive to natural enemies) and food odors have been found to be important in host-searching behavior of several species and will probably be found to be important in others. Learning to associate a food odor with the presence of its host has been observed in one parasitoid species, and this ability is likely to be important in others. The effect of temperature and relative humidity on the ability of a natural enemy to control its host has been determined for several parasitoids and predators. This suggests that biological control is compatible and complementary to recommended aeration procedures in grain bins. ELISA immunological assays that are capable of detecting and quantifying insect contamination in grain products have been developed for two parasitoids. Toxicological studies to measure the sensitivity of parasitoids to insecticides are just beginning.

Computer models are being developed to simulate parasitoid-host population dynamics of principal pests and parasitoids in bulk-stored wheat and corn, but this work needs to be extended to include additional species. Medium-scale experiments to determine appropriate release frequencies and rates necessary to control different pest species are being performed under a range of environmental conditions, but this work needs to be expanded to other species and to full-scale warehouse and bin storage situations. Releases of natural enemies have been evaluated in commercial peanut warehouses. Comprehensive pilot tests to evaluate biological control in wheat, corn and rice bins at Savannah, Manhattan and Beaumont have begun. These pilot tests are designed to 1) determine the efficacy of biological control agents for suppressing pest populations and maintaining grain quality, 2) evaluate application rates and release protocols, and 3) assess the impact of the release of beneficial insects on insect fragment counts in milled grain.

Pathogens. Strains of Bacillus thuringiensis (Bt) have been shown to control several moth pests, including Indianmeal moth and almond moth, in a variety of stored grains. Studies to find strains active against Indianmeal moth and navel orange worm are in early stages on nuts and dried fruits. Studies to find strains active against beetles in stored grain and tobacco have been encouraging, but have not reached commercialization. Extensive studies have demonstrated stability of Bt and granulosis virus (GV) in storage environments. These microbial insecticides are compatible with other control strategies and are not toxic to parasitoids or predators. This makes them ideal for use in integrated pest management (IPM) programs. Laboratory studies have shown the potential of two moth pests to evolve resistance to the Bt toxin. Potential for resistance also has recently been confirmed in several field crop pests. These findings raise questions about the long-term durability of Bt when used as a conventional insecticide and in transgenic plant applications. As a result, resistance management strategies are needed. A granulosis virus (GV) has been extensively studied to control moths in nuts and dried fruits. A patent has been awarded for the production and formulation of this GV strain. Preliminary studies to evaluate cytoplasmic (CPV) and nuclear polyhedrosis viruses (NPV) to control moths in stored nuts are ongoing. Protozoan pathogens have been found in a wide range of stored product pests, including beetles, but they have not been extensively evaluated for control.

Host Plant Resistance. It is widely recognized that plant varietal differences in chemical and physical attributes affect storage pests, but this has not been exploited to any significant extent. Recent advances in gene transfer technology now make it possible to introduce genes from other species into plants to make them resistant to pests. Transgenic walnuts and tobacco containing Bt toxin are being evaluated for the control of storage pests. Protease and amylase inhibitors of digestive enzymes occurring in wheat varieties are being evaluated for their effect on development rate, fecundity and survivorship of weevils, flour beetles and other species.

Biological Insecticides. Biological insecticides are naturally-occurring chemical compounds found in living organisms that could be used as toxins or repellents to control pest populations. Linalool, an extract from mint plants, is being evaluated as a stored grain insecticide and fumigant. Defensive volatile compounds from predatory bugs are being characterized and evaluated as repellents or toxins.

RESEARCH PRIORITIES

Parasitoids and Predators

- Biological studies - Demographic, behavioral, genetic, physiological, semiochemical, and ecological studies to provide basic information on efficacy and specificity of natural enemies that attack pests in bulk-stored grain and warehouses.
- Simulation models - Develop and validate computer models of natural enemy-pest population dynamics to predict and optimize pest suppression under different environmental conditions.
- Large scale testing - Release natural enemies in commercial-scale grain bins and warehouses to evaluate efficacy of direct and space treatments, and test release protocols.
- Mass-production - Nutritional studies and culturing techniques are needed to develop artificial hosts, reduce costs, and maintain quality of mass-reared biological control agents. Development of criteria and methods for assessing the quality of biological control agents will permit standardization crucial to commercialization.

- Behavioral and genetic modification - Learning behavior can be used to improve the host-searching efficiency of biological control agents by conditioning them to the appropriate host and food odors before release. Genetic selection may improve effectiveness by reducing susceptibility to insecticides, improving parasitism and host-killing efficiency, or decreasing dispersal.
- Determine the interactions between insect natural enemies and pathogens of their hosts. Are the effects additive, synergistic, or agonistic?

Pathogens (*Bacillus thuringiensis*, Viruses, Protozoa, Fungi)

- Efficacy/specificity - Examine mechanisms of specificity and develop new isolates, strains and formulations with more emphasis on major beetle pests.
- Formulation - Develop formulation, production, storage, and quality-control technologies.
- Genetic engineering - Develop novel deployment methods and modify host spectrum and virulence.
- Integrated pest management - Develop IPM programs incorporating strategies to minimize pest resistance to microbial insecticides. Research in this system will have implications for resistance management in crop production where insecticides or transgenic plants expressing toxins are used.

Host plant resistance

- Traditional plant breeding - Develop plant varieties expressing amylase or protease digestive enzyme inhibitors or physical characteristics that increase resistance to insect damage.
- Transgenic plants - Develop and evaluate stored products expressing Bt toxins or enzyme inhibitors.

Biologicals (toxins or repellents found naturally-occurring in living organisms)

- Identify and evaluate novel chemical compounds for toxicity and repellency.

Appendix A

USDA-ARS WORKSHOP ON STORED PRODUCT INSECTS Savannah, Georgia May 4-6, 1992

AGENDA

May 4 (Monday)

- 4:00 - Until Registration
6:00 - 7:00 Mixer
7:00 - Until Buffet Dinner

May 5 (Tuesday) AM

Moderator: Wendell Burkholder

- 7:00 - 8:00 Late Registration
8:00 - 8:15 Opening Remarks

Invited Speakers

- 8:15 Robert Richardson, Manager, Inspection Services, General Mills Inc.
Minneapolis, MN - "Processing Perspectives on Stored Product Pests"
- 8:55 Gary Obenauf, Director of Research, California Prune, Raisin and
Walnut Research, Fresno, CA - "Dried Fruit and Nut Industries"
- 9:35 Henry Bahn, USDA, S&E, Extension Service, Pioneer Hi-Bred
International Inc., Johnson, IA - "A Seed Company Perspective and
Relationship with Stored Product Insects"
- 11:10 Michael Fitzner, USDA, S&E, Extension Service, Washington, DC -
"Stored Product Integrated Pest Management: Risk Driven Practices"
- 11:50 Shlomo Navarro, Department of Stored Products, AGO, The Volcani
Center, Bet Dagen, Israel - "Research Priorities on Stored Product
Protection in Israel"
- 12:30 Lunch

May 5 (Tuesday) PM

Moderator: Patrick Vail

- 1:30 - 3:00 I Physiology, biochemistry, cell biology and genetics
*Organizers: Karl Kramer, Paul Shirk, James Baker
- 3:00 - 3:30 II Ecology, behavior, population biology, pheromone/attractant
discovery and development
*Organizers: James Throne, Thomas Phillips, David Hagstrum
- 3:30 - 3:45 Break
- 3:45 - 4:45 II Continued

May 6 (Wednesday) AM

Moderator: Richard Arbogast

- 8:00 - 9:30 III Detection, sampling, IPM systems
*Organizers: Paul Flinn, James Coffelt, Michael Mullen
- 9:30 - 10:00 IV Chemical and physical control
*Organizers: Larry Zettler, Patrick Vail, James Leesch
- 10:00 - 10:15 Break
- 10:15 - 11:15 IV Continued
- 11:15 - 11:45 V Biological Control
*Organizers: Lincoln Smith, William McGaughey,
Richard Arbogast
- 11:45 - 1:00 Lunch

May 6 (Wednesday) PM

Moderator: William McGaughey

- 1:00 - 2:00 V Continued
- 2:00 - 4:30 Panel discussion by industry and action agencies,
"Problems and Research Priorities"
- 4:30 - 5:00 Closing remarks, National Program Staff, ARS, USDA

*Lead Organizers are underlined.

Appendix B

Stored-Product Insects Workshop Participants 4-6 May 1992 Savannah, Georgia

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Appendix C

CURRENT RESEARCH INFORMATION SYSTEM (CRIS) PROJECTS

<u>Location/Scientist</u>	<u>CRIS No./Title</u>
<u>Beaumont, TX</u>	
Cogburn	6203-43000-002-00D, Bionomics and Control of Stored Product Insects in Rice.
	05000-0002-021-00 (Biocontrol Pilot Test). Suppression of Insect Populations in Stored Grain by Augmentation of Parasites and Predators.
<u>Fresno, TX</u>	
Curtis	5302-43000-019-00D, Integrated Pest Management and Basic Biology for Codling Moth on Walnut and Nectarine in Storage
Johnson	5302-53000-017-00D, Physical Control; Controlled Atmosphere, Heat and Cold for Dried Fruit, Nut and Nectarine Pests.
Soderstrom	5302-43000-017-00D, Physical Control; Controller Atmosphere, Heat and Cold for Dried Fruit, Nut and Nectarine Pests.
Vail	5302-43000-018-00D, NPV, Bacillus thuringiensis, Nematodes to Reduce Postharvest Insect Losses to Fruits and Nuts.
<u>Gainesville, FL</u>	
Coffelt	6615-43000-005-00D, Population Surveillance and Disruption of Reproduction of Stored-Product Insects.
Oberlander, Silhacek	6615-43000-004-02R, Biorational Suppression of Lepidopteran Pests of Field Crops and Stored Products.

Shirk	6615-43000-005-00D, Population Surveillance and Disruption of Reproduction of Stored-Product Insects.
Shuman	6615-43000-006-00D, Acoustical Communication and Electronic Detection of Insect Populations.
Silhacek	6615-43000-004-02R, Biorational Suppression of Shirk Lepidopteram Pests of Field Crops and Stored Products.
Silhacek, Oberlander	6615-43000-004-00D, Bioregulation of Growth and Development of Stored Product Insects.
<u>Madison, WI</u>	
Burkholder	3655-43000-002-00D, Using Ecology, Behavior, and Biochemistry for Biorational Management of Stored Product Insects.
	3655-43000-002-06S, Development of Rapid Biochemical Methods for Determining Insect Contamination in Grain Products.
Phillips	3655-43000-002-00D, Using Ecology, Behavior and Biochemistry for Biorational Management of Stored Product Insects.
	3655-43000-002-04T, Semiochemicals Across Trophic Levels: Moths and Parasitoids in Stored Products.
<u>Manhattan, KS</u>	
Beeman	5430-43000-008-00D, Development of Biological, Physiological and Genetic Controls of Stored Product Insects.
	5430-43000-010-00D, Integrated Insect Control Systems for the Stored Grain Industry.

Dowdy	<p>5430-43000-009-00D, Prediction of Insect Population Growth and Survival in Stored Grain Systems.</p> <p>5430-43000-011-00D, Biologically-based IPM Systems for the Grain Marketing and Processing Industry.</p>
Flinn	<p>5430-43000-009-00D, Prediction of Insect Population Growth and Survival in Stored Grain Systems.</p> <p>5430-43000-010-00D, Integrated Insect Control Systems for the Stored Grain Industry.</p> <p>0500-00001-026 (ARS Pilot Test), Expert System for Management of Insect Pests of Stored Wheat.</p> <p>0500-00002-020 (ARS Pilot Test), Suppression of Insect Populations in Stored Grain by Augmentation of Parasites.</p>
Hagstrum	<p>5430-43000-009-00D, Prediction of Insect Population Growth and Survival in Stored Grain Systems.</p> <p>0500-00001-004-00 (ARS Pilot Test), Automation of Stored Grain Insect Population Monitoring with Acoustical Sensors.</p>
Howard	<p>5430-43000-008-00D, Development of Biological, Physiological and Genetic Controls of Stored Product Insects.</p> <p>5430-43000-009-00D, Prediction of Insect Population Growth and Survival in Stored Grain Systems.</p>
Johnson	<p>5430-43000-008-00D, Development of Biological, Physiological and Genetic Controls of Stored Product Insects.</p> <p>5430-43000-010-00D, Integrated Insect Control Systems for the Stored Grain Industry.</p>
Kramer	<p>5430-43000-008-00D, Development of Biological, Physiological and Genetic Controls of Stored Product Insects.</p>

5430-43000-010-00D, Integrated Insect Control Systems for the Stored Grain Industry.

5430-43000-008-05T (USDA Competitive Grant), Pest Control by Manipulation of Insect Cytinolytic Enzymes and Their Genes.

McGaughey

5430-43000-009-00D, Prediction of Insect Population Growth and Survival in Stored Grain Systems.

5430-43000-010-00D, Integrated Insect Control Systems for the Stored Grain Industry.

5430-43000-011-00D, Biologically-based IPM Systems for the Grain Marketing and Processing Industry.

Oxford, NC

Keever

6647-43000-002-00D, Biological and Chemical Control of Insects that Damage Tobacco and Other Agricultural Products in Storage.

Savannah, GA

Arbogast

6605-43640-017-00D, Population Ecology and Predictive Models for Bulk Commodities.

Arthur

6605-43000-021-00D, Integrated Pest Management Strategies for Stored Product Insects.

6605-43000-011-021T, Develop Technology Related to the Application of Pesticides as an Insect Control Intervention in Warehouses.

Baker

6605-43000-020-00D, Biological Control of Storage Pests with Parasitoids and Predators.

Brower

6605-43000-020-00D, Biological Control of Storage Pests with Parasitoids and Predators.

0500-00002-018-00D, Suppression of Insect Populations in Stored Grain by Augmentation of Parasite and Predators.

Cline	6605-43640-017-00D, Population Ecology and Predictive Models for Bulk Commodities.
Leesch	6605-43000-019-00D, New Disinfestation Technologies for Stored Product Pests.
Mullen, Press, Wileyto	6605-43640-018-00D, Population Ecology and Predictive Models for Packaged Commodities.
Simonaitis	6605-43000-019-00D, New Disinfestation Technologies for Stored Product Pests.
Smith	6605-43640-017-00D, Population Ecology and Predictive Models for Bulk Commodities.
Zettler	6605-43000-021-00D, Integrated Pest Management Strategies for Stored Product Insects.

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